Effects of Plant Growth Regulators on Sturdiness of Jack Pine Seedlings[1]

Jeffrey P. Schnurr[2], Zong-Ming Cheng[3] and Arthur A. Boe[4]
Department of Plant Sciences
North Dakota State University, Fargo, ND 58105

Abstract

Seven growth regulators (ancymidol, 6-benzylaminopurine, ethephon, flurprimidol, mefluidide, paclobutrazol, and uniconizole) at various concentrations were studied for their effects on sturdiness of greenhouse grown seedlings of jack pine (*Pinus banksiana* Lamb.), measured by a single parameter, growth modification index. All growth regulators at certain concentrations were effective in reducing plant height but none increased stem caliper. Several growth regulators increased shoot-to-root dry weight ratio. Benzylaminopurine greatly modified growth characteristics and resulted in deformed seedlings. Uniconizole was most promising for producing sturdier seedlings of jack pine.

Index words: conifer, growth retardants, nursery crop production, *Pinus banksiana* Lamb.

Growth regulators used in this study: A-Rest (ancymidol), α-cyclopropyl-α-(4-methoxyphenyl)-5-pyrimidinemethanol; BAP, 6-benzylaminopurine; Cutless (flurprimidol), 1-(4-trifluoromethoxybenzyl)-1-pyrimidine-2-methyl-1-propanol; Bonzi (paclobutrazol), (2RS,3RS)-1-(4-chlorophenyl)-4-4-dimethyl-2-(1,2,4-triazol-1-yl)-pentan-3-ol; Embark (mefluidide), N-(2,4-dimethyl-5-(trifluoromethylsulfonylamino)-phenylacetamide; Florel (ethephon), (2-chloroethyl) phosphonic acid; Sumagic (uniconizole), (E)-(p-chlorophenyl)-4,4-dimethyl-2-(1,2,4-triazol-1-yl)-1-penten-3-ol.

Significance to the Nursery Industry

Tree seedlings spaced too close together in the nursery often produce tall, weak trunks that do not stand erect in the harsh field environment. These seedlings grow too quick in height and do not achieve adequate caliper growth, consequently, they do not establish and survive well in shelterbelt and windbreak plantings due to biotic and abiotic stresses (2). The results in this paper suggest that the application of certain growth regulators to the seedlings in the nursery may influence the growth characteristics such that they would be sturdier and more suitable for transplanting into the field. A single parameter, the growth modification index, was also established to measure the effect of a growth regulator on sturdiness of a seedling.

Introduction

A problem in the production of pines for windbreak and shelterbelt plantings is that high density nursery plantings result in production of tall and spindly seedlings. For example, Towner State Nursery in North Dakota produces jack pine (*Pinus banksiana* Lamb.) seedlings in stylefoam blocks and ships them as containerized stock for windbreak plantings. The transplanted pines often bend over during manual irrigation and may become embedded in the soil during heavy rain or strong wind. This problem can be avoided by planting seedlings with a stronger trunk. Several growth regulators have been used to enhance caliper growth, and may be useful for modifying jack pine seedling morphology. Application of 6-benzylaminopurine (BAP) at 200–600 mg/l to *Pinus sylvestris* and *P. nigra* seedlings resulted in short, highly branched plants (1). A chemical closely related to ancymidol, EL-500, was shown to reduce the height of 2-year-old *P. taeda* and *P. elliottii* var. *elliottii* seedlings (3). Tomato transplant spindliness due to high planting density was reduced by an ethephone spray (4). Several chemicals applied to *Pseudotsuga menziesii* var. *menziesii*, *P. taeda*, *P. coulteri*, *Cupressu arizonica*, and *Sequoia sempervirens* had variable effects upon seedling growth (5). Foliage application of paclobutrazol at 5 g/l significantly inhibited seedling growth of *Picea glauca* and *Pseudotsuga menziesii* (7). It was also reported that paclobutrazol, when used as a soil drench at 1–10 mg per pot, reduced the seedling growth of both *Pseudotsuga menziesii* var. *menziesii*, *P. taeda* (9).

The purpose of this study was to investigate the effects of several growth regulators on growth and development of young jack pine seedlings to produce less spindly plants. More specifically, we wanted to determine if there were chemicals that could produce seedlings with stronger trunks.

Materials and Methods

Seeds of jack pine were sown, on March 1, in 3.6 × 21.1 cm (1.4 × 8.25 in) Leach tubes held in 98 cell trays, a routine practice of seedling production in greenhouse. After each tube was filled to just below the top with Jiffy Mix (Jiffy Products of America, Inc., West Chicago, IL), two seeds were placed in each tube. About an additional 1 cm (~1/2 in) of Jiffy Mix was added and the contents were tamped lightly. The tubes were watered, about once per week, with fertilizer provided through an injecting system. The fertilizer was Peters 20–10–20 Peat-Lite Special containing supplemental micronutrients (Scotts Testing Laboratory, Allentown, PA). The seedlings were grown at 25/16C (77/61F), day/night, and in a 16-hr photoperiod. The light was provided by metal halide bulbs (150 $\mu mol \cdot m^{-2} \cdot s^{-1}$ at bench level) and the temperature was controlled with evaporative coolers. These conditions were maintained throughout the experiment. The greenhouse was fumigated with insecticides twice a week.

One month after germination, the less vigorous seedling in each tube was removed. For the experimental treatments, uniform plants were selected from the seedling population. The experiment was a randomized complete block design

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[2]Research Specialist II. Current address: Department of Forest Resources, University of Idaho, Moscow, Idaho 83844.
[3]Assistant Professor.
[4]Professor Emeritus. Current address: 1125 NE 1 St., Faribault, MN 55021.

(RCBD) with a total of 28 treatments, including four replications with 7 seedlings per treatment. A total of 784 seedlings were used in the experiment.

Seedlings were treated with growth regulators 90 days after sowing. A hand sprayer was used to apply the chemicals until the foliage was completely wetted. Each chemical was mixed with distilled water containing 1% dimethyl sulfoxide (DMSO) (Sigma, St. Louis, MO) and 0.1% Tween 20 (Sigma) surfactant before application. Water treatment was used as the control. All treatments used in this study are listed in Table 1.

Seedling height, caliper, shoot dry weight, and root dry weight were measured 120 days after the treatments were applied. Height was measured from the soil surface to the tip of the elongating leader. Caliper was measured about 1 cm (0.4 in) above the soil surface. A height : caliper ratio was also determined. The dry weight was a measurement of three seedlings randomly selected from each of four replications, or a total of 12 plants. Shoots and roots of a seedling were divided at the soil surface. The Jiffy Mix was carefully removed from roots under running water. Both shoot and root dry weights were determined after desiccation at 45C (113F) in a drying oven for three days. A shoot:root dry weight ratio was calculated using the individual values.

For the ratios of shoot height:caliper and shoot:root dry weights, a value of relative change was calculated using the control ratio of the water treatment. The formula was: relative change = [(control ratio – treatment ratio)/control ratio] × 100. For each treatment, a growth modification index was obtained by adding the two values of relative changes, since both characteristics were considered equally important. A higher index value indicates a sturdier tree with a heavier root system.

The two-way analysis of variance (ANOVA) for RCBD was done with SAS version 5 (6). The observations of seedling height, caliper, shoot dry weight, root dry weight, and the two calculated ratios were subjected to ANOVA. Mean separation was done with the least significant difference (LSD) method where there was no missing data ($P \leq 0.05$). Where the design was unbalanced due to missing data, means were separated by probability of difference ($P \leq 0.05$) (6).

Results and Discussion

The effects of growth regulators on primary growth parameters are shown in Table 1. DMSO, the solvent for growth regulators, did not affect growth of jack pine seedlings in comparing with the water control. This suggests that the effects of growth regulators are not due to the solvent they were dissolved in. All growth regulators at most concentrations tested, significantly reduced shoot height and stem caliper in comparing with the water control, with BAP at 400 mg/l causing most severe height reduction (Table 1). BAP has also been shown to reduce seedling heights in *Pinus sylvestris*, and *P. nigra* seedlings at 200–600 mg/l (1). PAC also significantly inhibited seedling growth of *Picea glauca*

Table 1. Effect of foliar sprays of several growth regulators on growth of *Pinus banksiana*.

Chemical	Concentration (mg/l)	Shoot height (cm)[z]	Stem caliper (cm)	Height/ caliper ratio	Dry weight (DR)(g) shoot	Dry weight (DR)(g) root	Height: caliper ratio	H:C relative change[y]	S:R relative change[x]	Growth modification index[w]
Water		18.9	0.37	50.4	2.77	1.97	1.41	–	–	–
Water + DMSO		19.5	0.38	52.1	3.07	2.09	1.4	–3.4	–4.3	–7.7
Benzylaminopurine (BAP)	50	14.8*	0.31*	48.5	1.87*	1.06*	1.76	3.8	–24.8	–21.0
	100	14.7*	0.32*	45.9	1.65*	0.86*	1.92*	8.9	–36.2	–27.3
	200	10.1*	0.26*	38.7*	1.24*	0.61*	2.03*	23.2	–44.0	–20.8
	400	8.3*	0.23*	36.6*	0.75*	0.31*	2.29*	27.4	–71.6	–44.2
Paclobutrazol (PAC)	10	18.1	0.36	50.2	2.62	1.70	1.54	0.4	–9.2	–8.8
	25	14.0*	0.31*	44.4	2.13	1.58	1.35	11.9	4.3	16.2
	50	16.2	0.33*	49.1	2.41	1.70	1.42	2.6	–0.7	1.9
	100	13.2*	0.31*	42.0	2.26	1.96	1.15	16.7	18.4	35.1
Uniconizole (UNI)	10	13.8*	0.31*	44.9	1.83*	1.42	1.29	10.9	8.5	19.4
	25	12.7*	0.30*	43.0	2.27	1.88	1.21	14.7	14.2	28.9
	50	11.8*	0.29*	40.8*	2.11	1.81	1.17	19.0	17.0	36.0
	100	11.8*	0.31*	38.1*	2.22	2.10	1.06	24.4	24.8	49.2
Flurprimidol (FLU)	20	13.7*	0.31*	43.9	2.36	1.69	1.40	12.9	0.7	13.6
	50	14.5*	0.30*	49.3	2.06	1.55	1.33	2.3	5.7	8.0
	100	14.8*	0.29*	50.7	1.97*	1.62	1.22	–0.6	13.5	12.9
	200	11.8*	0.27*	43.3	1.91*	1.62	1.18	14.1	16.3	30.4
Ancymidol (ANC)	20	16.6	0.32*	51.4	2.78	1.95	1.43	–2.0	1.4	–0.6
	50	15.4	0.31*	49.6	1.75*	1.22*	1.43	1.6	1.4	3.0
	100	14.4*	0.31*	45.9	2.92	2.32	1.26	8.9	10.6	19.5
	200	12.6*	0.32*	39.9*	2.27	1.86	1.22	20.8	13.5	34.3
Ethephon (ETH)	1000	15.8	0.34	46.1	2.25	1.57	1.43	8.5	1.4	9.9
	2000	12.6*	0.30*	42.1	1.77*	1.21*	1.46	16.5	–3.5	13.0
Mefluidide (MEF)	20	16.0	0.34	47.6	2.47	1.79	1.38	5.5	2.1	7.6
	50	17.8	0.31*	58.3	2.19	1.18*	1.86*	–15.7	–31.9	–47.6
	100	17.7	0.31*	57.8	2.56	1.47	1.74	–14.7	–23.4	–38.1
	200	13.9*	0.27*	58.8	1.99	1.34	1.49	–16.7	–5.7	–22.4

[z]* Significantly different from the water control. Mean separation with probability of difference ($P \leq 0.05$).

[y]H:C relative change: height:caliper relative change = [(control ratio – treatment ratio)/control ratio] × 100.

[x]S:R relative change: shoot:root relative change = [(control ratio – treatment ratio)/control ratio] × 100.

[w]Growth modification index = (height:caliper relative change) + (shoot:root relative change).

and *Pseudotsuga menziesii* when applied to foliage at 5 g/l (7). Five treatments, which caused a significant decrease in shoot height:stem caliper ratios, were BAP at 200 and 400 mg/l, UNI at 50 and 100 mg/l, and ANC at 200 mg/l (Table 1). This is probably due to decrease in both shoot height and stem caliper in most treatments (Table 1).

Seedling sturdiness can be affected by both ratios of shoot height to stem caliper, and the ratio of shoot-to-root dry weight, although most previously reported research only described changes in shoot height:caliper (1, 3, 4, 5, 7, 9). Decrease of shoot-to-root dry weight ratio should result in sturdier seedlings which is more likely to survive the stresses related to windbreak and shelterbelt plantings, because plants can anchor better and establish faster with the large root system. In this experiment, shoot and root dry weights were affected by fewer growth regulators than shoot height and stem caliper (Table 1). BAP at all four concentrations incited a large decrease of shoot and root dry weights. Other treatments which caused a decrease of shoot dry weight included ANC at 50 mg/l, ETH at 2000 mg/l, UNI at 10 mg/l, and FLU at 100 and 200 mg/l. Besides BAP, three other treatments, ANC at 50 mg/l, ETH at 2000 mg/l, and MEF at 50 mg/l, caused significant reduction of root dry weight (Table 1). Four treatments increased the ratio of shoot-to-root dry weight of which three were BAP treatments (Table 1). No treatments caused a significant decrease in the shoot-to-root dry weight ratio in comparing with the water control (Table 1).

To determine which treatments produced the most sturdy plants, it is difficult to reach a conclusion by examining individual parameters and/or ratios. For example, the best treatment in comparing to water control for reducing the shoot height:caliper ratio (BAP at 400 mg/l) also caused the most undesirable increase of the shoot:root dry weight (Table 1) and growth deformation. To determine the overall effect of a chemical on the seedling sturdiness, we developed a growth modification index by adapting the simple index method used to select for multiple traits during plant breeding (10). To generate such an index value, relative changes of both ratios were calculated to eliminate unit differences between height/caliper and weight, then, these two values are added together, to yield a growth modification index (Table 1, last column). An index value below zero indicates a more negative effect on sturdiness by the chemical, and a value above zero indicates a positive effect on sturdier growth.

With this single index, the effects of different chemicals and concentrations can be determined easily by comparing their index values. For example, the BAP at 50–400 mg/l caused positive changes, or a decrease in the ratios of height to caliper, but this was more than offset by a very large undesirable change, or an increase in the ratios of shoot-to-root dry weights. These treatments have index values far below zero, ranging from –20.8 to –44.2 (Table 1). These were among the poorest treatments and the plants were severely deformed and unhealthy. Another set of treatments which had large negative index values were MEF at 50–200 mg/l (Table 1). MEF did not cause major deformation like BAP, but the plants were taller and more spindly than the water control plants, and were not desirable. The treatments with the highest index values included UNI at 50 and 100, PAC at 100, FLU at 200, and ANC at 200 mg/l. These treatments were also the best by visual observations with a good root system, but reduced heights (data not shown). Overall, UNI was the best for inducing more sturdy, healthy plants, as all four treatments had high index values (Table 1). The same plant growth regulator at different concentrations showed different effects, as indicated by the index values (Table 1).

Forest nurseries often produce spindly conifer seedlings due to too close spacing in the nursery or in the greenhouse, and these plants are not very suitable for windbreak planting due to harsh environmental conditions. Shorter plants with stronger trunks will be beneficial. We reported here the positive effect of a number of growth regulators on sturdiness of jack pine seedlings. Our results generally agree with those previous reports in a number of other species (1, 3, 4, 8), such that BAP induces short, highly branched plants with less developed root system (1). Seedling sturdiness can be significantly increased by a single spray of UNI at 10 to 100 mg/l. These results should be applicable to nurseries which produce jack pine or other conifer plants for windbreak or other uses. The simple index value developed to quantitatively measure the effect of growth regulators on sturdiness can also be used in other species, or in similar situations.

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